REMARKS

Claims 39-68 currently appear in this application. The Office Action of March 9, 2004, has been carefully studied. These claims define novel and unobvious subject matter under Sections 102 and 103 of 35 U.S.C., and therefore should be allowed. Applicants respectfully request favorable reconsideration, entry of the present amendment, and formal allowance of the claims.

Specification

The specification is objected to because it contains an embedded hyperlink.

Accordingly, this hyperlink has been deleted from the specification.

Claim Objections

Claims 13-17 are objected to because of the term "genetical modification." Claim 14 is objected to because the word "heterologous" is misspelled in line 3 of the claim.

Claims 1-28 have now been replaced by claims 39-68. It is believed that the new claims contain no informalities.

Rejections under 35 U.S.C. 101

Claims 34 and 35 are rejected under 35 U.S.C. 101 because the claimed invention is directed to non-statutory subject matter, in that they read on naturally occurring animal embryos and animals.

Claims 34 and 35 have been cancelled. Therefore, this rejection is now moot.

Rejections under 35 U.S.C. 112

Claims 1-38 are rejected under 35 U.S.C. 112, first paragraph, because the Examiner alleges that the specification is only enabling for reconstructing an ovine embryo comprising transferring a G1 or G0 diploid donor ovine cell into an enucleated metaphase II ovine donor cell or a G1 or G0 diploid donor ovine cells into an enucleated metaphase II ovine oocyte, wherein the chromatin of the donor ovine nucleus is subjected to denaturing conditions before transfer into the recipient ovine oocyte, activating the resulting nuclear transfer unit, culturing the resulting nuclear transfer unit to form blastocysts, transferring the blastocysts into an ovine surrogate mother and allowing the blastocysts to develop to term to form an ovine.

This rejection is respectfully traversed. The present invention does not pretend to teach nuclear transfer methods, which were well known at the time the present application was filed. The present invention is directed to a further step to apply this known method in order to maximize the number of successful implants. As is evident from the documents cited by the Examiner, the NT procedure in various different species was well known at the time of filing of the

present application as well as of the priority date of the present application (see Dinnyes et al., "Somatic Cell Nuclear Transfer: Recent Progresses and Challenges," *Cloning and Stem Cells* **4(1)**: 2002, page 82, column 1, "cloning in various species," where it is expressly said that nuclear transfer on sheep (1996), cattle (1998), goats (1999) and mice (1998) have been successfully produced. Attention is also directed to Westhusin et al, "Cloning to Produce Desired Genotypes," page 36, lines 12-24, in which cloning through nuclear transfer is reported for mice, cattle goats, sheep, with references cited that are dated well before the priority date of the present application.

The rationale of the present invention lies, as specified on page 12, lines 30-34 of the specification as filed, in that successful cloning through nuclear transfer techniques may be jeopardized by the organization of chromatin. The difficulties in reprogramming chromatin, a target that is still a difficult task (see Dinnyes et al., "Somatic Cell Nuclear Transfer: Recent Progresses and Challenges," *Cloning and Stem Cells* **4(1)**: 2002, page 86, column 2 "main problems limiting efficiency", last three lines, page 87, column 1, lines 1-5, etc.) and often the rate of success in cloning procedures has been low.

The present application solves the problem by providing a preliminary step to add to conventional nuclear transfer protocols that leads to a very good reprogramming of chromatin, and hence to a higher yield of successful development of embryos obtained by nuclear transfer techniques.

The added step of the present invention involves denaturing the chromatin of the donor cell prior to nuclear transfer. In fact, Applicant has surprisingly discovered that denaturation of nuclear chromatin prior to NT does not cause defects in the subsequent NT procedure and subsequent embryo growth, but, on the contrary, permits improved programming of eth chromatin, leading to a higher yield of success in the subsequent embryonic growth and development.

Although nuclear transfer protocols are quite species-specific, as the Examiner has noted, many successful protocols suitable for different animal species were known in the state of the art before the priority date of the present application. The object of the invention is not an entirely new protocol for NT, but an improvement in conventional NT protocol. This improvement involves denaturing the chromatin of the donor cell prior to transfer of the nucleus. The technical effect of this step is to produce a greater yield of *in vitro* cultured embryos obtained with respect to the

standard protocols known in the art (see Example s and Table 1, pages 20-22 of the specification), as well as a higher yield in the development of embryos once they are transferred into recipient animals, particularly in the first stages of development (see Table 2 of the specification), with a pregnancy rate of 80-100% in contrast to the 50% obtained without the denaturation step.

Because of the high conservation of the chromatin structure and regulation systems in all eukaryotes, this step is clearly applicable to all species with the same result of permitting better reprogramming of chromatin.

The above also applies to the rejection regarding the improving step when applied to nuclear transfer in producing transgenic animals. Again, the present application does not pretend to teach techniques for constructing transgenic animals, but rather teaches a novel step preceding nuclear transfer that greatly increase the success in the cloning procedure because of successful reprogramming of chromatin that follows the denaturation step.

Furthermore, although the choice of the expression cassette is crucial when producing a transgenic animal, it is also true that one skilled in the art would have found, at the time of filing the present application, a good number of suitable expression cassettes in the scientific literature,

for the species disclosed in the present specification. This is well documented in the papers the Examiner cited, demonstrating that, at the time of filing the present application, successful creating of transgenic sheep, goats, mice, calves, and other animals were well known in the art.

The process of the present invention can also be used when cloning existing transgenic animals, wherein the choice of the expression cassette and the successful positioning of the cassette is already well known.

Therefore, it is respectfully requested that the rejection of the claims based upon lack of enablement be withdrawn because, by the priority date of the present application various protocols for NT in different species, as well as suitable expression cassettes for different species, were available to those skilled in the art. As shown in Table 2 of the present specification, the application of the novel denaturation step prior to nuclear transfer significantly increases the success of the technique.

With respect to the inter-species problem posed by the Examiner, although at the time of filing the present application various studies on inter-species techniques were already available, the specification merely points out that the efficiency of possible interspecies nuclear transfers will

be improved by the denaturing process of the present invention.

Claims 1, 2, 21, 22 and 24 are rejected under 35 U.S.C. 112, second paragraph, as being indefinite.

This rejection is respectfully traversed. Claims 1-38 have now been replaced with new claims 39-68. It is believed that these new claims clearly define the invention for which patent protection is sought. It has been made clear in the new claims that the denaturing step can be performed on the whole donor cell, the denaturing treatment being sufficient to denature the chromatin also when the nucleus is located inside the donor cells (as from Example 1, heat treatment, the cell indeed suffers some denaturation as well, but the nucleus is still well usable for the nuclear transfer, as disclosed on page 22, lines 9-12), or on the nucleus *per se*, once the nucleus is outside the donor cell. The new claims are worded so as not to extend beyond the wording of the specification as filed, although it is obviously implicit in the meaning of the specification that, in order to denature the nucleus outside the donor cell, the nucleus is previously extracted form the donor cell.

Art Rejections

Claim 35 is rejected under 35 U.S.C. 102(b) as being anticipated by Blakely et al.

Claim 35 has been cancelled and has not been replaced by a new claim. Therefore, this rejection is moot.

Claims 34 and 35 are rejected under 35 U.S.C. 102(b) as being anticipated by Gomez et al.

Claims 34 and 35 have been cancelled and have not been replaced by new claims. Therefore, this rejection is moot.

Claims 36-38 are rejected under 35 U.S.C. 102(b) as being anticipated by Schieke et al.

Claims 36-38 have been cancelled and have not been replaced by a new claim. Therefore, this rejection is moot.

In view of the above, it is respectfully submitted that the claims are now in condition for allowance, and favorable action thereon is earnestly solicited.

Respectfully submitted,

BROWDY AND NEIMARK, P.L.L.C.
Attorneys for Applicant

By _____
Anne M. Kornbau
Registration No. 25,884

AMK:msp
Telephone No.: (202) 628-5197
Facsimile No.: (202) 737-3528
G:\BN\S\socf\Loi1\PTO\21may04.amd.doc